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A Further Note on a Case of Dermal Leishmaniasis from S. America, with the Results of Inoculation Experiments.

(Successful Inoculation of a Cat.)

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In a former number of this Journal (Vol. I. Pt. 3) I described a case of dermal leishmaniasis from S. America. The case was one of an Englishman who had contracted the disease in Peru or Bolivia, where he had been travelling. He had two sores—one a large one over the right eye, and another smaller one at the outer corner of the same eye. The present note deals with the subsequent history of this case after its treatment was commenced, the result of the culture of the leishmania obtained from it, and some inoculation experiments on animals.

Treatment.—A reference to my former paper will show that the case was treated in two ways. In the first place, killed cultures of the leishmania were injected subcutaneously. This treatment did not appear to be checking the disease, as the smaller sore showed signs of spreading towards the eye. Accordingly a second mode of treatment was adopted in the form of applications of an ointment made of equal parts of methylene-blue, lanoline, and vaseline, as recommended by Cardamatis and Melissidis. The ointment was applied on a piece of lint, the size of the sore, both morning and evening. This caused very little pain, at any rate nothing that the patient was unwilling to endure. The effect of the ointment was to cause a great increase in the exudation, which was of liquid consistency and of a dark blue colour. The ointment was applied to the small sore at the outer corner of the eye for two weeks, and, as it showed signs of healing, the applications were stopped. In another fortnight healing was complete. To the larger sore over the eye treatment with the ointment was continued for two and a half months, and, as it was thought that possibly the ointment had killed the leishmania but was preventing healing, the treatment was stopped and a simple ointment applied. In two weeks' time the sore had completely healed. Since then the patient has been seen from time to time. He was last seen ten months after the sores had healed, and there was no sign of any recurrence. The glandular enlargement, which was such a marked feature of the case, had steadily diminished and had practically disappeared, though one of the nodules under the skin of the right eyebrow could still be detected, though it was much reduced in size. The resultant scarring was very slight compared with the extent of the lesions, and this would have been still less had not an attempt been made to excise the larger sore before the patient came under treatment and the condition had been diagnosed.

It is to be hoped that this treatment with the methylene-blue ointment will be given a good trial, for in the present case it has had a very pleasing result.

Inoculation Experiments with the Cultures of the Parasite.

In the earlier paper I described how a culture of the leishmania was obtained on N. N. N. medium. The growth in this medium was very luxuriant, so that it was easy to maintain the culture by subinoculating tubes. This was successfully carried out for seventeen generations in N. N. N. medium, so that there is every probability that, as with other strains of leishmania, it would be possible to carry on the culture indefinitely. Compared with a culture of *Leishmania tropica*, obtained from the Oriental Sore of Aleppo, the South American leishmania grew much more readily, though morphologically no constant differences could be made out between the two types of parasite.

With the cultures some inoculation experiments were made :—

1. With the first culture obtained (Nicolle's culture virus) a human being was inoculated on the skin of the forearm in two places—one by intracutaneous injection with a hypodermic needle, and the other by scarification. A year has now elapsed since the inoculation and no infection has taken place.

2. A young rabbit was inoculated intravenously on two occasions with rich cultures of the leishmania, but no infection took place.

3. A young rabbit was inoculated intravenously on one occasion with rich culture, but did not become infected.

4. Attempts were also made to infect mice by intraperitoneal injection of rich cultures, but without success.

Experiments with the Virus from the Sore.

I previously described the successful inoculation of a dog and baboon directly from the sores on the man :—

1. The baboon was inoculated in the skin on each side of the nose and on the tips of the ears by means of a hypodermic syringe. After an incubation-period of four months, sores containing leishmania had appeared on the ears and nose. The sore on the nose was in the form of a papule which persisted for about two months and finally disappeared without ulceration. The lesions on the ears were in the form of small papules which ultimately reached the size of a pea, and, owing probably to scratching by the monkey, finally ulcerated. They persisted for six months, and then gradually disappeared, finally healing at the end of the seventh month. Shortly after this the monkey died from some unknown cause, but no sign of leishmania could be found in its organs.

On August 22, 1912, a black dog was inoculated on each ear in several places with material obtained by scraping one of the sores on the baboon. After twenty-three days three small papules had appeared on the dog's left ear. In these leishmania were discovered. The incubation-period was thus much shorter than in the case of the baboon inoculated directly from the man. Later nodules appeared on the other ear. These increased slightly in size, but had disappeared in five months.

2. In my former paper I recorded the successful inoculation of a dog (i) directly from the man. A sore developed on each ear after an incubation of two months. One sore was excised for inoculation purposes, the other was left. Though seven months have elapsed since its appearance, it has not yet disappeared, but is now represented by a shallow ulcer. The excised sore was emulsified with saline solution and injected into each ear of another dog (ii). After an incubation-period of one month dog (ii) had developed a small nodule on one ear, and two weeks later a similar nodule on the other ear. One month later one of these nodules was excised and emulsified with saline for inoculating other animals (*a*, *b*, *c*). The remaining one was left alone. It persisted for three months and then absorbed. From the emulsion of the sore the following animals (*a*, *b*, *c*) were inoculated :—

a. Small puppy intravenously. The animal, previously healthy, began to lose weight and had a temperature up to 102·1° F. during the next month. It was in such poor condition that a general infection was suspected. During this time, however, liver puncture (twice) and bone marrow puncture (once) gave only a negative result as regards leishmania. Two months after inoculation, the dog appeared to be improving in health, so was killed. Smears from its organs were negative, as were also six tubes of N. N. N. medium inoculated from its spleen.

b. Two mice inoculated intraperitoneally did not become infected.

c. A cat inoculated in each ear developed small pinhead nodules after two months at the sites of inoculation. These have increased slightly in size, and now, five months after inoculation, are still present, while leishmania have been discovered in them first by the culture method on N. N. N. medium, and later by microscopical examination of smears from the sore.

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